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Changing rainfall frequency rather than drought rapidly alters annual soil respiration in a tropical forest

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ABSTRACT

Tropical forests play an important role in global carbon (C) cycling due to high primary productivity and rapid litter and soil organic C decomposition. However, it is still unclear how changing rainfall will influence soil CO_2 losses (i.e. via soil respiration) in tropical forests. Here, using a rainfall and litter manipulation experiment in a tropical forest, we show that enhanced litter-leached dissolved organic carbon (DOC) production with increased rainfall frequency drives substantial CO_2 loss via soil respiration. A 50% increase in rainfall frequency (no change in total rainfall amount) enhanced inputs of DOC by 28%, total dissolved nitrogen (TDN) by 17%, and total dissolved phosphorus (TDP) by 34% through leaching from litter layer to soil surface likely due to faster litter decomposition rate, and stimulated soil respiration by ~17% (about 1.16 t C ha^{-1} yr^{-1}). Soil respiration responded to altered rainfall frequency with limited when litter layer was removed. Accordingly, soil microbial biomass C (MBC) and fine root biomass were increased by 23% and 20%, respectively only in the plots with litter layer. A 50% reduction in total rainfall (no change in rainfall frequency) did not change litter-leached DOC and nutrients fluxes, soil MBC, fine root biomass, or annual mean soil respiration rates. The new finding – that enhanced leached-DOC production with increased rainfall frequency drives profound increases in soil respiration in tropical forests – suggests that future climate changes may have significant impacts on soil C dynamics and global C budget, and argues for the importance of incorporating this underappreciated feedback into prognostic models used to predict future C-climate interactions.

1. Introduction

Tropical forests are a critical component of the global carbon (C) cycle. They contain approximately 20% of the global soil C stock and account for roughly 35% of terrestrial net primary productivity (Jobbagy and Jackson, 2000). Responses of tropical forest C dynamics to climate change could significantly influence atmospheric carbon dioxide (CO_2) concentrations and thus affect the pace of future climate change (Houghton et al., 2015). Climate models generally predict more drought events and changes in rainfall frequency around the tropics in the future (Zhou et al., 2011; IPCC, 2013), with potentially profound effects on tropical forest productivity and tree mortality (Doughty et al., 2014). However, the effects of shifting rainfall patterns on tropical forest soil CO_2 fluxes (e.g., via soil respiration) and the underlying mechanisms are still not fully understood (Knapp et al., 2008; Bond-Lamberty and Thompson, 2010). As a result, model predictions of soil respiration in tropical forests were often inconsistent with experimental observations, making the impact of potential changes in rainfall patterns on the tropical forest C balance highly uncertain (Powell, 2014).

Both field experiments and model simulations have identified soil temperature and soil moisture as two important controls on soil respiration (Davidson and Janssens, 2006; Falloon et al., 2011). Changing rainfall can affect soil respiration mainly via altered soil moisture (Fig. 1a; Gabriel and Kellman, 2014; Vicca et al., 2014; Liu et al., 2016). Indeed, rainfall-manipulation experiments in temperate ecosystems mostly show declines in soil respiration in response to simulated drought and increases in soil respiration with simulated increases in rainfall (Wu et al., 2011; Liu et al., 2016). However, the paucity of studies conducted to date in tropical forests have produced different results (Sotta et al., 2007; Davidson et al., 2008; Cleveland et al., 2010;

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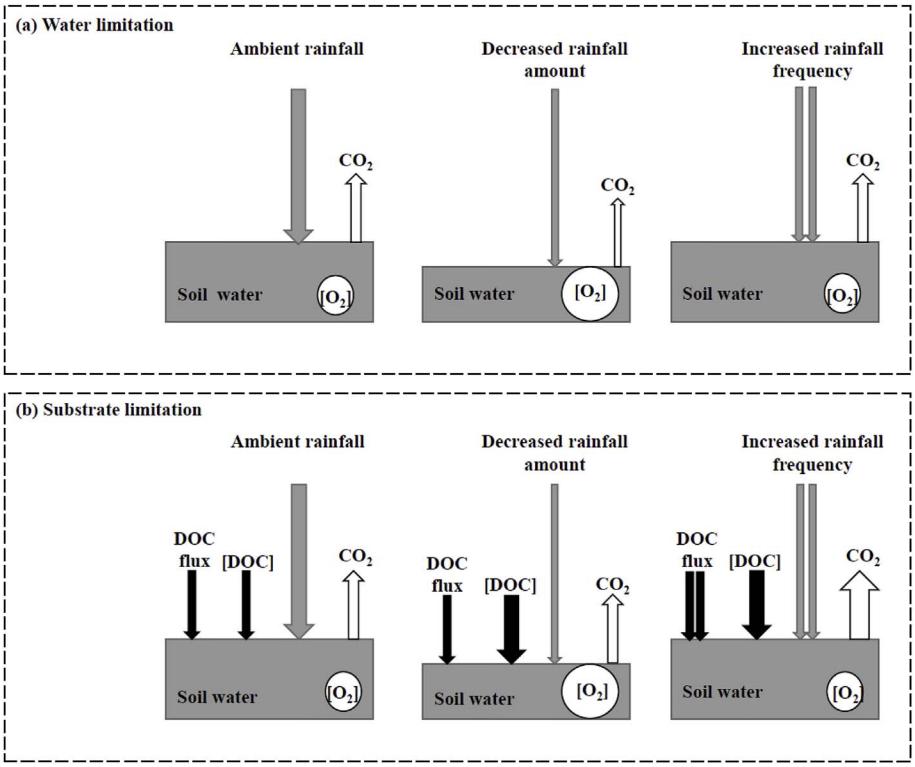

Fig. 1. Conceptual models illustrating the potential effects of rainfall changes on soil respiration. a) A traditional model that includes the trade-off between soil water and O_2 concentration ($[O_2]$). b) A revised model (Cleveland et al., 2010) that integrates the potential effects of litter-leached dissolved organic carbon (DOC) concentration ([DOC]) and DOC flux into the traditional model. Our revised model includes the potential effects of the mechanistic controls from water limitation to substrate limitation. The sizes and the number of the arrows or lettering reflect relative differences in response to rainfall changes.

Van Straaten et al., 2010; Deng et al., 2012; Wood and Silver, 2012), with inconsistencies often attributed to a trade-off between soil water content and soil O_2 concentrations (Fig. 1a).

In addition to soil temperature and soil moisture, C substrate (quality and quantity) also strongly regulates rates of soil respiration (Davidson and Janssens, 2006; Xu et al., 2014; Campbell et al., 2016). Studies using rainfall simulations or laboratory incubations have demonstrated that pulses of rainfall can transport large quantities of dissolved organic carbon (DOC) from the litter layer, and leached DOC fluxes can stimulate large episodic CO_2 pulses from the soil surface (Cleveland et al., 2007; Wu and Lee, 2011). Given the consistently warm temperatures and ample rainfall in many tropical forests, frequent litter-leached DOC inputs may have disproportionately strong effects on soil respiration, although the intensity of such effects may depend on the quality of C input and the status of soil organic matter in the study sites (Cleveland et al., 2007; Qiao et al., 2014; Zhou et al., 2016). Thus, rainfall changes may significantly affect tropical forest soil respiration by altering annual input of litter-leached DOC. Cleveland et al. (2010) showed that experimental drought did not change total amount of litter-leached DOC flux, but significantly enhanced its concentration, and stimulated CO_2 fluxes from soils. Change in rainfall *frequency* has been shown to exert a greater influence on litter decomposition rates than changes in total rainfall amount (Vanlauwe et al., 1995; Wieder et al., 2009; Anaya et al., 2012), implying that altered rainfall frequency could have more profound effects on litter-leached DOC fluxes and soil respiration rates (Fig. 1b). Unfortunately, few, if any, field experiments have explicitly manipulated rainfall frequency in a tropical forest ecosystem, severely limiting our ability to predict how climate change-driven shifts in rainfall patterns may alter the C cycle in this important biome (Beier et al., 2012; Liu et al., 2016).

We conducted a one-year rainfall manipulation experiment in an old-growth tropical forest southern China (23°10′ N and 112°10′ E). Three rainfall treatments were chosen: 1) an ambient rainfall as a control (CK); 2) a 50% increase in rainfall frequency with no change in total rainfall amount (increased rainfall frequency treatment; IRF); and 3) a 50% reduction of rainfall amount with no change in rainfall frequency (drought treatment; DRA) (Fig. S1). We hypothesized that the DRA treatment would decrease soil respiration due to reduced soil moisture (Fig. 1a), while the IRF treatment would increase soil respiration due to enhanced input in litter-leached DOC flux (Fig. 1b). To test this hypothesis, we further quantified the contributions of different soil CO_2 sources, and specifically isolated the indirect soil CO_2 flux driven by litter-leached DOC (R_{DOC}) through either permanently or temporally removing litter layer (See more details in the methods). We expected that the IRF treatment would increase R_{DOC}, without significant changes in the other soil CO_2 sources.

2. Materials and methods

2.1. Experimental design

This study was carried out in an old-growth tropical forest at the Dinghushan Biosphere Reserve (DBR) in the Guangdong Province in southern China (23°10′ N and 112°10′ E). The forest is dominated by *Castanopsis chinensis, Cryptocarya concinna, Schima superba, Machilus chinensis*. No disturbances were recorded for the past 400 years in this forest (Zhou et al., 2016). Soil properties and major stand information of this tropical forest have been shown in Table S1. Climate is typical south subtropical monsoon climate, with mean annual temperature of 21.4 °C, and mean annual precipitation of 1956 mm, of which nearly 80% falls in the hot-humid wet/rainy season (April–September) and 20% in the cool-dry season (October–March). However, long-term observation records in this region showed that rainfall frequency, intensity and seasonal patterns have been highly variable for the past three decades (Zhou et al., 2011).

A randomized block design with three blocks was established in June 2013. The rainfall treatments were randomly assigned to plots in each block. Each plot was $5 \times 10\ m^2$, and the distance between plots was more than 5 m. A 5-m PVC panel was inserted in the middle of the plot to divide each plot into two subplots ($5 \times 5\ m^2$). One subplot was used for litter removal treatment and another subplot received normal litter fall.

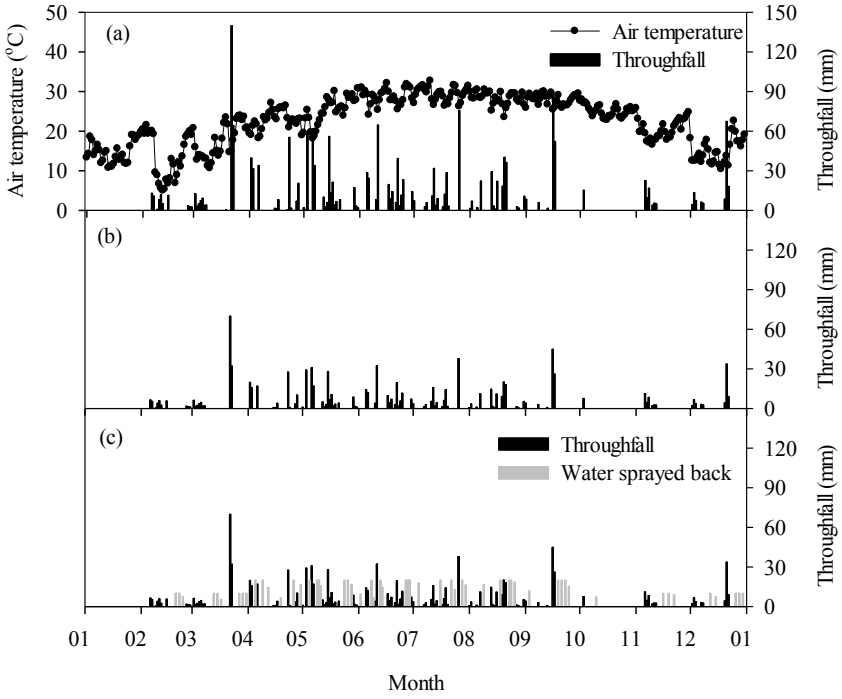

Fig. 2. Daily air temperature and throughfall under the canopy of the control CK (a), DRA (b) and IRF (c) plots. The rainfall treatments are: DRA = decreased rainfall amount, CK = control, IRF = increased rainfall frequency. Error bars represent ± 1 standard errors.

The rainfall treatments included a control (ambient rainfall that is from natural rain events; CK) and two altered rainfall treatments: 1) a 50% increase in rainfall frequency with no change in total rainfall amount (IRF); and 2) a 50% reduction in rainfall amount with no change in rainfall frequency (DRA) (Fig. S1). For the DRA treatment (Fig. S1c), a 50% of the total throughfall was intercepted using semi-circle and transparent PVC panels (10-cm diameter). The intercepted throughfall from each plot was transferred to two big buckets for determination. For the IRF treatment (Fig. S1d), half of the throughfall was also intercepted and collected in the buckets, but re-applied to the plots to simulate the increase in rainfall frequency without a change in total rainfall amount. The water in the buckets was sprayed back evenly using a backpack sprayer when the collected throughfall was over 5 mm and 4–5 days after the rain in the dry season or 1–2 days in the wet season. If the collected throughfall was > 10 mm in the dry season or > 20 mm in the wet season due to sustained rainy days or individual extreme rainfall event, it was re-applied in multiple events with 10 mm water per application event during the dry season and 20 mm per application event in the wet season. Canopy throughfall was monitored using a Hobo Micro Station (H21-002, Onset Computer Corporation, Pocasset, USA) near the experimental plots.

2.2. Measurements

Twelve PVC soil collars were permanently installed in each plot, with four in each "litter removal" subplot and eight in each "litter intact" subplot (Fig. S2a). Soil respiration rates were measured twice a month in 2014 from four soil collars in the "litter intact" subplots (R_{total}) (Fig. S2c) and four soil collars in the "litter removal" subplots (R_{bare}) (Fig. S2b), respectively using a Li-Cor 8100 Soil CO_2 Flux system (Li-Cor Inc., Lincoln, NE, USA) equipped with a survey chamber. The difference between R_{total} and R_{bare} represents the total contribution of litter to soil respiration. To isolate the direct CO_2 release from litter layer (R_{litter}) and the indirect CO_2 release from soil driven by litter-leached DOC (R_{DOC}), we further measured soil respiration in the "litter intact" subplots ($R_{no\text{-}litter}$) (Fig. S2d) by temporally removing litter from the other four of the eight collars in the "litter intact" subplots before 1–2 h of the measurement. The removed litter in such collars was then added back after measurement (Fig. S2e). Thus, the R_{litter} was calculated as the difference between R_{total} and $R_{no\text{-}litter}$. The R_{DOC} was calculated as the difference between $R_{no\text{-}litter}$ and R_{bare}.

To quantify dissolved organic carbon (DOC) delivery from the litter layer to the soil surface, each of the litter subplots was equipped with four stainless steel dishes (400 cm^2) placed under the litter layer. Each stainless steel dish was covered with a 0.5-mm mesh nylon screen to exclude large debris. The litter-leached DOC was intercepted by the stainless steel dish, and flowed to a plastic bottle through a small plastic pipe. The leached volume in the plastic bottle was determined after each rainfall event and a subsample from each bottle was collected two times per month. The collected leachate was immediately frozen for subsequent C and nutrient analyses. The concentrations of DOC and total dissolved nitrogen (TDN) in the samples were determined using a Shimadzu TOC analyzer (TOC-VCPH, Shimadzu, Japan), and total dissolved phosphorus (TDP) was analyzed colorimetrically (Anderson and Ingram, 1989).

Soil microbial biomass C (MBC) and fine root biomass (diameter ≤ 2 mm) at 0–20 cm depth were determined every three months. Four samples of twelve cores were randomly collected from each subplot each sampling. The soil MBC was calculated using the fumigation–extraction method. The fine roots were separated by washing and sieving, dried at 60 °C for 48 h and weighed.

2.3. Statistical analysis

Replicate measurements were averaged by subplot for each sampling day before the statistical analysis. Since soil temperature or moisture between the "litter intact" subplots and the "litter removal" subplots was not significantly different (*t*-test, $p > 0.05$ for both) for each rainfall treatment, we averaged them in the two different litter subplots. A repeated measures Analyses of Variance (RM-ANOVA) was performed using daily mean to test the difference of soil respiration (including all CO_2 sources), soil temperature, soil moisture, litter-leached DOC concentration and flux by rainfall treatment and season. Multiple comparisons (Least Significant Difference, LSD method) were

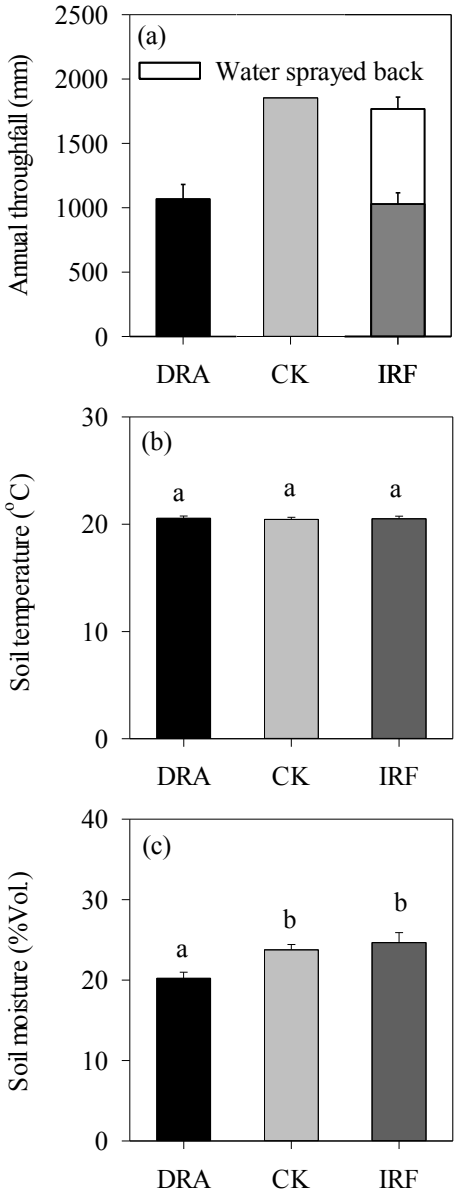

Fig. 3. Annual mean values of throughfall (a), soil temperature (b), and soil moisture (b) under different rainfall treatments. The rainfall treatments are: DRM = decreased rainfall amount, CK = control, IRF = increased rainfall frequency. Error bars are standard errors. Mean values in each season denote significant difference ($p < 0.05$) among rainfall treatments.

conducted if significant effects of rainfall treatment or season were found. Similar RM-ANOVAs were also performed to test the difference of fine root and soil microbial biomass by rainfall treatment and season for the "litter intact" subplots and the "litter removal" subplots, respectively.

3. Results

3.1. Rainfall manipulation

In total, the control plots received 1854.8 mm of throughfall in 108 days (Figs. 2a and 3a), of which nearly 71% fell in the hot-humid wet/rainy season (April–September) and 29% in the cool-dry season (October–March). The DRA plots on average received 1067.8 mm of throughfall (Figs. 2b and 3a), representing a 42% reduction in total throughfall relative to the control plots. In the IRF plots, 1029 .5 mm of throughfall was captured (Figs. 2c and 3a) and 778.2 mm of intercepted

Table 1
Significance of the effects of rainfall treatment, season and their interactions on soil respiration rate from all CO_2 sources was tested using Analysis of Variance (ANOVA). The CO_2 sources are: R_{total} = total soil respiration, R_{bare} = bare soil respiration, R_{litte} = litter respiration, R_{DOC} = DOC-driven soil respiration. Numbers are F-values. Stars indicate the level of significance ($^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$). DRA = decreased rainfall amount, IRF = increased rainfall frequency.

	Soil respiration rate			
	R_{total}	R_{bare}	R_{litte}	R_{DOC}
Rainfall	17.17***	1.18	60.47***	31.72***
Season	246.37***	107.86***	108.70***	300.65***
Rainfall × Season	2.60	3.57*	2.08	13.27***

throughfall was sprayed back to the IRF plots for 55 time (days) (Figs. 2c and 3a), resulting in a total of 1767.7 mm throughfall in the plots (Fig. 3a), similar to the control plots. Thus, re-applying the captured throughfall to the IRF plots increased 51% throughfall events (rainfall frequency) annually compared to the control (Fig. 2a, c, 3a).

3.2. Soil microclimatic factors

The seasonality of soil temperature and moisture was consistent with the seasonal patterns of air temperature and throughfall, with higher values in the wet season than in the dry season ($P < 0.01$ for all) (Table 1). Litter treatment did not significantly alter soil temperature or moisture in all three rainfall treatment ($p > 0.05$ for all). In the control plots, annual mean soil temperature is 20.45 ± 0.03 °C, and annual mean soil moisture is 23.75 ± 0.09 %Vol (Fig. 3b and c). Soil temperature was not changed by the rainfall treatments ($P > 0.05$; Table 1 and Fig. 3b). Soil moisture was significantly decreased by 28% under the DRA treatment compared to the control ($P < 0.001$; Table 1 and Fig. 3c). No significant change in soil moisture was detected between in the IRF treatment and in the control ($P > 0.05$; Table 1 and Fig. 3c).

3.3. Soil respiration rate

Soil respiration rate of all CO_2 sources also exhibited a clear seasonal pattern with the maximum respiration rates occurred during the wet season, whereas the minimum respiration rates occurred during the dry season ($P < 0.01$ for all) (Table 1 and Fig. 4a, c, e, g). Annual mean soil respiration rates in the control plots were estimated as 2.67 ± 0.09 for R_{total}, 1.60 ± 0.09 for R_{bare}, 0.56 ± 0.03 for R_{litter}, and 0.51 ± 0.0.3 for R_{DOC} (Fig. 4b, d, f, h). R_{total} was significantly stimulated by ~17% under the IRF treatment compared to the control ($P < 0.001$) (Table 1 and Fig. 4b). Both R_{bare} and R_{litter} were not changed by the IRF treatment ($P > 0.05$) (Fig. 4d, f). The estimated R_{DOC} increased by 67% under the IRF treatment compared to the control ($P < 0.001$) (Table 1 and Fig. 4h). By contrast, only the R_{litter} was significantly decreased by 32% under the DRA treatment compared to the control ($P < 0.001$) (Table 1 and Fig. 4f). For the other CO_2 sources, no significant change was detected between the DRA treatment and the control ($P > 0.05$) (Table 1 and Fig. 4d, h). The effect of rainfall treatments on soil respiration varied between seasons (Table 1). The IRF treatment increased R_{DOC} primarily in the wet season (Fig. 4g). By contrast, the DRA treatment decreased soil respiration of all CO_2 sources in the dry season, but not in the wet season (Fig. 4a, c, e, g).

3.4. Litter-leached dissolved organic matter

The concentration of litter-leached DOC and nutrients did not change between seasons ($P > 0.05$ for all), while the litter-leached DOC and nutrients fluxes were significantly higher in the wet season than in the dry season ($P < 0.01$ for all). In the control plots, annual

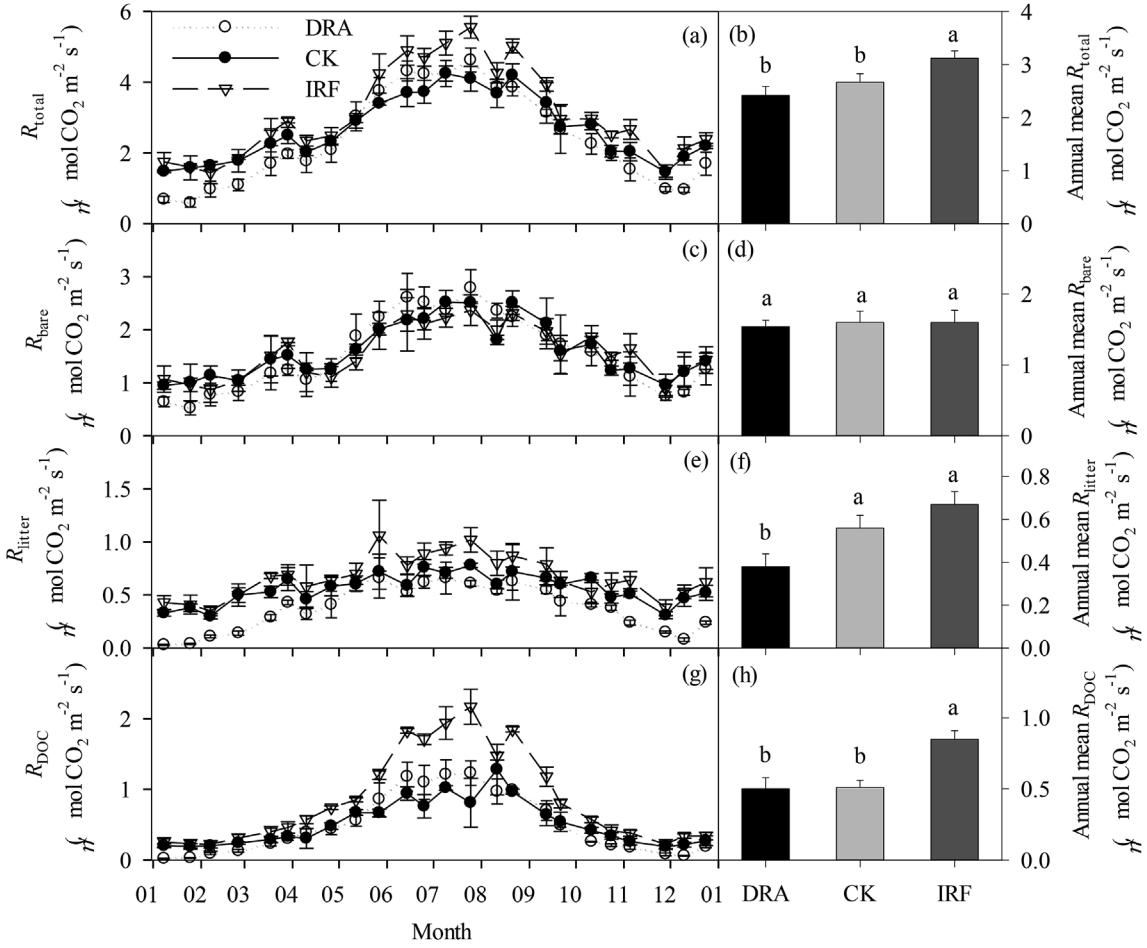

Fig. 4. Seasonal variations and annual mean values of total soil respiration (R_{total}, a and b), bare soil respiration (R_{bare}, c and d), litter respiration (R_{litte}, e and f), and DOC-driven soil respiration (R_{DOC}, g and h) under rainfall treatments. The rainfall treatments are: DRM = decreased rainfall amount, CK = control, IRF = increased rainfall frequency. Error bars are standard errors. Different letters above columns denote significant difference ($p < 0.05$) among rainfall treatments.

mean concentrations of litter-leached DOC, TDN, and TDP were 91.85 ± 3.11 mg L^{-1}, 2.58 ± 0.19 mg L^{-1} and 0.19 ± 0.02 mg L^{-1}, respectively. The mean fluxes of litter-leached DOC, TDN, and TDP were 249.50 ± 14.71 kg ha^{-1} yr^{-1}, 32.58 ± 3.20 kg C ha^{-1} yr^{-1}, and 2.63 ± 0.20 kg C ha^{-1} yr^{-1}, respectively (Fig. 5). The litter-leached DOC concentration was significantly increased by 36% under the DRA treatment compared to the control ($P < 0.001$; Table 1 and Fig. 5a), while total litter DOC flux did not change ($P > 0.05$; Fig. 5b). The litter-leached DOC concentration was significantly increased by 29% under the IRF treatment compared to the control ($P < 0.001$) (Table 1 and Fig. 5a), and total litter DOC flux was significantly increased by 28% ($P < 0.001$) (Table 1 and Fig. 5b). The concentration of litter-leached TDN was enhanced under the IRF treatment only, compared to the control (Fig. 5c). The flux of litter-leached TDN under the IRF treatment was significantly higher than that in the DRA treatment, but neither IRF nor DRA treatment showed significant difference in the flux of litter-leached TDN with the control (Fig. 5d). The concentrations of litter-leached TDP were enhanced by both DRA and IRF treatments (Fig. 5e), but only IFR treatment significantly increased the fluxes of litter-leached TDP by 34% (Fig. 5f).

3.5. Soil microbial biomass carbon and fine root biomass

Soil microbial biomass carbon (MBC) and fine root biomass was generally higher in the wet season than those in the dry season ($p < 0.05$ for both). The annual mean values of soil MBC and fine root biomass in the control were 134 g m^{-2} and 653 g kg^{-1} soil, respectively (Fig. 6). Litter removal significantly decreased soil MBC and fine root biomass in all three rainfall treatments ($p < 0.05$ for both). The DRM treatment did not change soil MBC and fine root biomass in both "litter intact" and "litter removal" subplots (Fig. 6). The IRF treatment significantly increased soil MBC by 23% and fine root biomass by 20% in the "litter intact" subplots, but not in the "litter removal" subplots (Fig. 6).

4. Discussion

The findings from our rainfall and litter manipulation experiment provide new insights into the effect of altered rainfall on soil respiration in tropical forests of southern China and may have significant implications for soil C dynamics and global C budget in future climate change. Overall, soil respiration responded strongly to altered rainfall frequency, but less to changes in rainfall amount (Fig. 4). This was consistent with previous observations in a subtropical forest in China, showing that rainfall frequency rather than annual rainfall amount change controls interannual variation of soil respiration (Wang et al., 2011). Annual mean R_{total} was significantly stimulated by ~17% (equivalent to 1.66 t C ha^{-1} yr^{-1}) under the IRF treatment compared to the control ($P < 0.001$; Table 1 and Fig. 4b), while R_{bare} showed no change between the two rainfall treatments ($P > 0.05$; Fig. 4d). The contrasting responses between the subplots with and without a litter layer indicate that the most tenable explanation of soil respiration increase under the IRF treatment lies in the processes occurring in the litter layer, rather than the effect of soil moisture or O_2 concentration

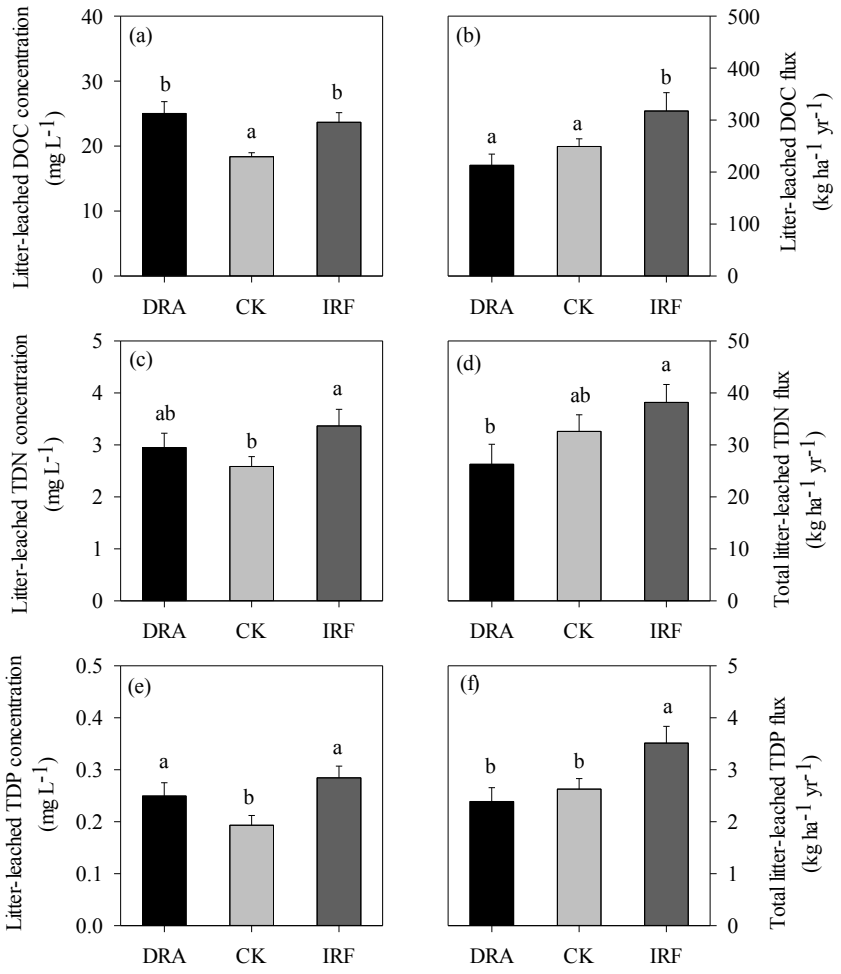

Fig. 5. Annual mean concentrations and fluxes of litter-leached dissolved organic carbon (DOC, a and b), total dissolved nitrogen (TDN, c and d), and total dissolved phosphorus (TDP, e and f) under different rainfall treatments. The rainfall treatments are: DRA = decreased rainfall amount, CK = control, IRF = increased rainfall frequency. Error bars represent ± 1 standard errors. Different letters above columns denote significant differences ($p < 0.05$) among rainfall treatments.

change. Moreover, neither soil moisture nor soil temperature change between the IRF treatment and the control was detected in both "litter intact" and "litter removal" subplots ($P > 0.05$ for both; Fig. 3b and c).

Consistent with our hypothesis, R_{litter} were also not changed by the IRF treatment ($P > 0.05$; Fig. 4f), while annual mean R_{DOC} was increased by 67% (equivalent to 1.28 t C ha^{-1} yr^{-1}) under the IRF treatment compared to the control ($P < 0.001$; Table 1 and Fig. 4h). Thus, we conclude that the increased R_{total} under the IRF treatment was primarily driven by the input of litter-leached DOC. Accordingly, in the IRF treatment the annual mean concentration of litter-leached DOC increased by 29% compared to the control ($P < 0.01$; Table 1 and Fig. 5a), leading to a 28% increases in net DOC fluxes ($P < 0.01$; Table 1 and Fig. 5b, f). The more input of litter-leached DOC could be attributed to faster litter decomposition rate with increasing rainfall frequency (Vanlauwe et al., 1995; Wieder et al., 2009; Anaya et al., 2012). A recent study in an Asian tropical rainforest also suggested that the weekly DOC flux passing through the hydrological processes (throughfall, litter leachate, soil water, and interception by the surface soil) significantly explained the dynamics of soil respiration rate, with higher sensitive indices than those for soil temperature and moisture (Zhou et al., 2016).

Several biological processes may help explain why an increase in

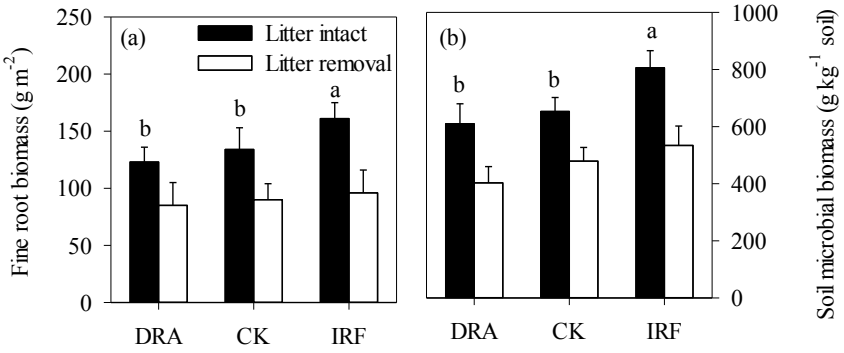

Fig. 6. Annual mean values of soil microbial biomass carbon (a) and fine root biomass (b) under different rainfall treatments. The rainfall treatments are: DRA = decreased rainfall amount, CK = control, IRF = increased rainfall frequency. Error bars are standard errors. Different letters above columns in the litter intact subplots denote significant difference ($p < 0.05$) among rainfall treatments. No significant difference in either fine root biomass or soil microbial biomass carbon among rainfall treatments was found in the litter removal subplots.

litter-leached DOC flux with increased rainfall frequency greatly enhanced soil respiration. First, elevated litter DOC fluxes could directly stimulate microbial respiration. Many studies have shown that labile C additions such as litter-leached DOC inputs rapidly stimulate microbial growth and respiration (Cleveland et al., 2007; de Graaff et al., 2010; Straathof et al., 2014), particularly in this old-growth tropical forest where a considerable portion of the soil organic C is non-readily oxidizable based on the $KMnO_4$ oxidation method (Chen et al., 2015; Table S1). Indeed, soil microbial biomass, an indicator of heterotrophic respiration, was significantly higher under the IRF treatment than the control when litter was not removed (Fig. 6b). Second, the enhanced total litter-leached DOC included a 34% additional increase in P compared to the control (Fig. 5f). Phosphorus has been shown to be a major factor limiting the plant productivity at our study site (Huang et al., 2012), and thus an increase in available P input via greater DOC flux may have stimulated plant and root growth. The increased fine root biomass in the "litter intact" subplots under the IRF treatment (Fig. 6a) indicated that root respiration could be enhanced (Wood and Silver, 2012). Finally, higher frequency and fluxes in litter-leached DOC could have indirectly stimulate microbial decomposition of old C previously storied in the soil, a phenomenon known as the "priming effect" (Sayer et al., 2011; Qiao et al., 2014; Canarini and Dijkstra, 2015).

It is notable that the response of soil respiration to the rainfall treatments varied among seasons (Table 1; Fig. 4). The lack of R_{DOC} response to the IRF treatment in the dry season was probably due to water limitation (Fig. 4g), as the input of litter DOC flux was raised as well. This is not surprising. Soil microbial activity might be inhibited during the dry season, in turn reducing the demand for C substrates. In contrast, the turnover rate of plant roots could be enhanced due to dry season-induced dieback, thus providing more labile C to cause soil microbes insensitive to litter-leached DOC. While the input of litter-leached DOC could benefit soil C accumulation in the dry season, it likely stimulates soil respiration in the following wet season when soil moisture return to the favor level. Future study should be conducted to explore the mechanisms underlying the seasonal differences of litter-leached DOC input impacts on soil respiration. Accordingly, soil respiration in the DRA treatment was significantly decreased in the dry season (Fig. 4a). This was consistent with previous studies in other tropical forests, showing that drought decreases soil respiration only during a natural dry period or year (Davidson et al., 2008; Van Straaten et al., 2010). The DRA treatment could significantly decrease R_{litter} (Fig. 4f). This was probably due to that the litter layer, being highly porous (porosity 90%) and directly exposed to canopy air, could not retain water for a long time and hence is usually at relatively low water potentials (typically < -50 MPa in non-rain days) even in wet forest ecosystems (Lee et al., 2004). Therefore, the litter decomposition rate and its CO_2 release in this tropical forest are still highly sensitive to rainfall changes particularly rainfall frequency change.

Our experiment is among the first to explicitly manipulate rainfall frequency change in a tropical forest, and shows that increasing rainfall frequency accelerates CO_2 losses via soil respiration. The findings from this experiment also provide new insights into the mechanistic controls of tropical forest soil respiration under rainfall changes. Soil respiration is mainly controlled by soil temperature, soil moisture and C substrate (Davidson and Janssens, 2006; Falloon et al., 2011; Xu et al., 2014; Campbell et al., 2016). Although soil temperature and moisture usually explained more the variations of soil respiration than C substrate, soil respiration is more sensitive to C substrate input that soil temperature and moisture. Several studies have shown that soil respiration rapidly increased with C addition, particularly for the input of litter-leached DOC (Cleveland et al., 2007; De Troyer et al., 2011; Kindler et al., 2011). By combining the litter removal treatment with a rainfall manipulation, we provide a direct, experimental evidence showing that the enhanced litter-leached DOC flux was the major driver of the increase in soil respiration with increased rainfall frequency (Fig. 1b). However, such a response is often ignored under experimental manipulation of rainfall amount due to its minor effect on litter-leached DOC flux. Our results indicated a highly complex hydrological DOC process, and this is the first study linking these processes with soil respiration and rainfall changes in tropical forests. The interaction between litter-leached DOC input and various rainfall patterns (including not only changing rainfall amount and frequency but also altering rainfall intensity) should be investigated in the future study. Incorporating these hydrological DOC processes into the soil respiration models may provide more consistent responses of soil respiration to rainfall changes in tropical forests.

More broadly, our study may actually provide a conservative picture of the effects of changing rainfall on soil respiration in tropical forests. For example, our site is characterized by a modest rainfall ($\sim$1950 mm y^{-1}) and a pronounced ($\sim$6 month) dry season, but our results of seasonal variations suggest the potential for greater impacts of altered rainfall frequency in wetter and/or more aseasonal tropical forests. However, other tropical forests may have various C storage and protection mechanisms in the soil and different vegetation composition that could alter the quality and quantity of litter-leached DOC. These factors could directly influence soil respiration responses, and should be further studied in the future. The long-term implications of rainfall change impacts on soil respiration could be complicated. Soil microbe may acclimatize to such greater input of litter-leached DOC over time. Plants may also change the ratios of root to shoot in response to shift of soil nutrient status. In addition, both changing rainfall frequency and drought alter the stoichiometry of litter-leached DOC, which may change the quality of soil organic matter in the near future and influence the soil respiration responses. Nonetheless, if the phenomena we observed here could apply generally to tropical forests, the enhanced litter-leached DOC flux with rainfall frequency change and its $\sim$18-fold potential to increase soil respiration revealed here would significantly influence both ecosystem C losses and the global climate system. Given this sizable response, some representation of these processes should be built into ecosystem models to provide more accurate estimation of the effects of climate-driven changes on the C cycle in tropical forests.

Statement of authorship

QD, DZ, QZ and DH designed the research. QD, XH and GC conducted the experiment and collected the experimental data. QD and D.H. performed the analysis. QD wrote the draft, and all co-authors edited the manuscript. The authors declare no competing financial interests.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.soilbio.2018.02.023.

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